

Figure 1 Qualitative Analysis of Cultured Chondrocyte Viability. Chondrocytes cultured on 2% FMC bipolymer (70/30) alginate scaffolds were grown in either 6mL of or 9mL of growth medium. After 7 days, cell viability was tested using a LIVE/DEAD kit. Live cells appear green and dead cells appear red. The cells in both samples were mainly viable, with a few dead cells found in each. An ImageJ analysis of these images counted 147 cells in the 6mL sample and 103 cells in the 9mL sample.

Sample	A_{260}/A_{280}	RNA Used in RT-PCR (ng in 20 μ L)
6 mL	1.6	10
9 mL	1.4	10

Table 1 RNA Spectrometry Analysis. RNA isolated from both 6 mL and 9 mL samples was scanned at A_{260} and A_{280} , and a purity ratio was calculated. From the absorbance values, RNA concentrations were calculated, and used to determine the amount of RNA run in RT-PCR.

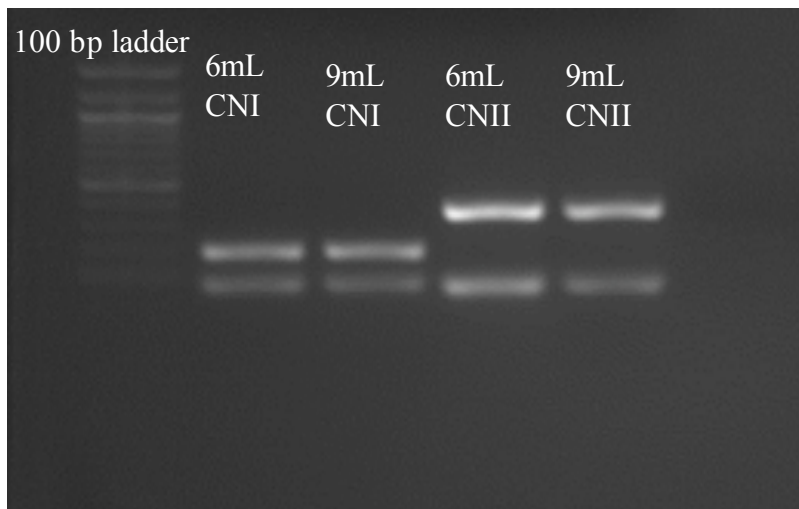


Figure 2 Qualitative Analysis of Collagen I and Collagen II in Experimental Chondrocyte Cultures. Using RT-PCR, cDNA was created from mRNA purified from chondrocyte cultures, amplified, and run on an agarose gel. The first lane indicates a DNA ladder, the next two lanes contains Collagen I cDNA from the 6 mL and 9mL samples, respectively. Both samples contain bright bands corresponding to Collagen I. The final two lanes contain Collagen II cDNA from the 6mL and 9mL samples, respectively. Both samples contain bright bands corresponding to Collagen II, with the 6mL sample band being slighter brighter. In all lanes, the bottom band represents a GAPDH loading control.

control CDR CNII/CNI Ratio:	1.23
experimental 6ml CDR CNII/CNI:	0.95
experimental 9ml CDR CNII/CNI:	1.13

Table 2 Semi-Qualitative RT-PCR Analysis of Collagen I and Collagen II Content in Experimental Samples. ImageJ analysis of RT-PCR gel seen in Figure 2 was preformed to analyze the amount of Collagen I and Collagen II cDNA in both the 6mL and 9mL samples. Control analysis of freshly isolated chondrocyte cells was also performed to provide a baseline for analysis. The chondrocytes grown in 9mL growth medium had a ratio closer to the control ratio than did those grown in 6mL medium.

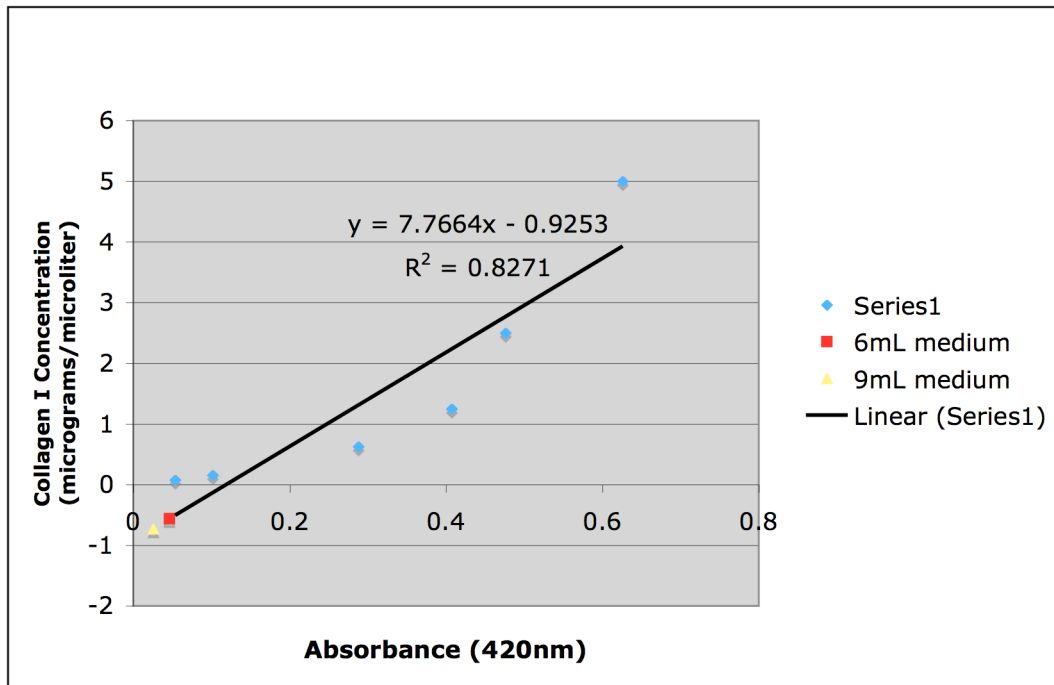


Figure 3. Results of ELISA Analysis to measure Collagen I concentration in Experimental Samples. Collagen I samples of known concentration were plotted against respective 420 nm absorbance values to create a standard curve. Experimental concentrations were calculated using the standard. In both the 6mL and 9mL samples, protein concentrations were below the detection limit.

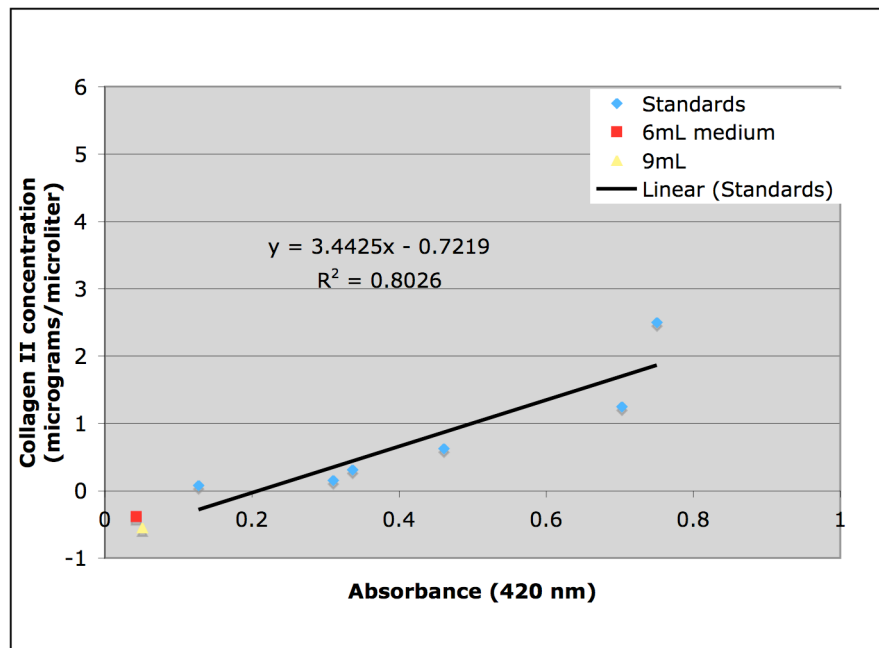


Figure 4. Results of ELISA Analysis to measure Collagen II concentration in Experimental Samples. An indirect ELISA analysis was performed on cell lysates from both the 9mL and 6mL experimental samples. Collagen II samples of known concentration were plotted against respective 420 nm absorbance values to create a

standard curve. Experimental concentrations were calculated using the standard. In both the 6mL and 9mL samples, protein concentrations were below the detection limit.

Sample	Collagen I concentration ($\mu\text{g}/\mu\text{l}$)	Collagen II concentration ($\mu\text{g}/\mu\text{l}$)
6mL CDR cell lysates	-.56	-.38
9mL CDR cell lysates	-.73	-.55

Table 3. Collagen I and II concentrations from ELISA analysis. An indirect ELISA analysis was performed on lysates from both the 9mL and 6mL experimental samples. Collagen I and II concentration was calculated from standard curves plotted from known solutions. (Figure 3+4).